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February 21, 1992

8EHQ-0292 - 2508 INIT

Document Processing Center (TS-790)
Office of Toxic Substances
Environmental Protection Agency
401 M Street, SW

Washington, D.C. 20460

Attention: Section 8(e) Coordinator (CAP Agreement)

Occidental Chemical Corporation ("OxyChem")

Toxic Substances Control Act ("TSCA") Section 8(e) Compliance Audit Program

Agreement No. 8ECAP-0063

Dear Sir:

Subject:

Attached find one original and two copies of the following document:

+ Aquatic Toxicity Hexachlorocyclopentadiene

This document is being submitted pursuant to the TSCA Section 8(e) Compliance Audit Program ("CAP") and a CAP agreement executed between OxyChem and the U. S. Environmental Protection Agency (Agreement No. 8ECAP-0063).

The identity of the chemical(s) tested in the study listed above are as follows:

Hexachlorocyclopentadiene, CAS# 77-47-4.

The adverse effect(s) noted in the study listed above are as follows:

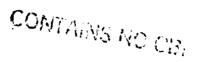
 Acute toxicity of <1 ppm at 96 hour under static exposure condition for several fish species; Bluegill Sunfish, Channel Catfish and the Fathead Minnow.

If you have any questions on the information contained herein, please contact me at (716) 286-3358.

 $\mathcal{O}^{\mathcal{O}}$

Ladd W. Smith

Director, Product Stewardship



ACUTE TOXICITY OF HEXACHLOROCYCLOPENTA-DIENE TO BLUEGILL (Lepomis macrochirus), CHANNEL CATFISH (Ictalurus punctatus), FATHEAD MINNOW (Pimephales promelas) AND THE WATER FLEA (Daphnia magna).

TOXICITY TEST REPORT

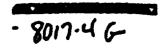
SUBMITTED TO

VELSICOL CHEMICAL CORPORATION

CHICAGO, ILLINOIS

E G & G, Bionomics

Aquatic Toxicology Laboratory
790 Main Street
Wareham, Massachusetts
March, 1977



INTRODUCTION

These investigations were performed at the Aquatic

Toxicology Laboratory of E G & G, Bionomics, Wareham,

Massachusetts to determine the susceptibility of bluegill

(Lepomis macrochirus), channel catfish (Ictalurus punctatus),

fathead minnow (Pimephales promelas) and the water flea

(Daphnia magna) to hexachlorocyclopentadiene in acute, 96-hour (fish) and 48-hour (water flea), toxicity tests.

MATERIALS AND METHODS

Hexachlorocyclopentadiene, an amber-colored liquid, tested on a product basis, was received from the Velsicol Chemical Corporation, Chicago, Illinois. Nominal test concentrations are reported as milligrams of hexachlorocyclopentadiene per liter of diluent water (mg/1).

Bluegill used in these toxicity tests were obtained from a commercial fish hatchery in Nebraska and had a mean and standard deviation (N=30) wet weight of 0.45 ± 0.17 grams (g) and standard length of 26 ± 4.9 millimeters (mm). Channel catfish were acquired from a fish hatchery in Missouri and had a mean and standard deviation (N=30) wet weight of 2.1 ± 1.0 g and a standard length of 54 ± 9.8 mm. Pathead minnow were acquired from a commercial fish hatchery in Arkansas and had a mean and standard deviation (N=30) wet weight of

 0.72 ± 0.3 g and standard length of 38 ± 5.2 mm. Upon receipt at Bionomics, fish were held in 1700-1 concrete raceways which were coated with an epoxy resin paint to prevent leaching of extranecus materials into the water. Well water with a hardness of 35 mg/L as CaCO,, a pH of 7.1, a temperature of 21 + 1.0°C and a dissolved oxygen (DO) concentration of greater than 60% of saturation flowed through these raceways at a minimum of 4 1/minute which provided an adequate water turnover rate for maintaining these species. Fish were maintained under these conditions for a minimum of 30 days prior to use in these tests. During the holding period, fish mortality was less than 2%. Fish were from the same year class and the standard length of the longest fish was no more than twice that of the shortest. Immediately prior to use in the tests, fish were acclimated to test conditions of temperature and water quality over a 48-hour period. Pish were not fed during the acclimation period, there was no mortality observed during this period and fish appeared to be in excellent condition at the initiation of the tests.

Water flea (< 24 hours old) used in this toxicity test were from laboratory stocks cultured at Bionomics. Well water used to culture these animals had the same water quality characteristics as the water previously described for the maintenance of raceway fish populations. Prior to use in the water flea test, diluent water was filtered through polyester fiber and

sterilized with an ultraviolet light to remove any microorganisms.

Procedures used in these acute toxicity tests were based on protocols in "Methods for Acute Toxicity Tests with Fish, Macroinvertebrates, and Amphibians" (U.S. EPA, 1975) except where stated otherwise.

The static fish toxicity tests were conducted in 19.6-liter glass jars which contained 15 liters of test solution. The test compound, dissolved in acetone, was added directly into each jar and mixed by stirring. The water flea toxicity test was conducted in 250 milliliter (ml) beakers which contained 150 mt of test solution. For each test concentration, the appropriate amount of the test compound, dissolved in acetone, was pipetted into 500 ml of diluent water and mixed with a magnetic stirrer. This solution was then divided into three equal aliquots in triplicate beakers to provide replicate exposure treatments. The remaining 50 mf were used for 0-hour pH and DO determinations. Diluent water used in both tests had the same water quality characteristics as described previously for holding water. Neg tive controls, which consisted of the same dilution water and conditions as test concentrations, but with no hexachlorocyclopentadiene or acetone, were established. In addition, positive controls containing the greatest amount of acetone introduced into any one test

vessel were also established. Test vessels were maintained in water baths at $22 \pm 1.0^{\circ}C$ and test solutions were not aerated during the tests. Ten bluegill and fathead minnow, and five channel catfish (10 animals per concentration) were randomly assigned to each test jar and five water flea (15 animals per concentration) were randomly assigned to each test beaker within 30 minutes after the compound was added.

Two independent tests involving two different series of concentrations were established in each toxicity test. The preliminary (range-finding) test defined a narrower range of concentrations to be used in the definitive test which then would assess the dose-response relationship and permit calculation of a median lethal concentration (LC50) and its 95% confidence interval from the mortality data produced.

During these tests, the dissolved oxygen concentration, pH and temperature of test solutions were checked at 0, 24, 48 and 96 hours of exposure for fish and 0 and 48 hours for water flea in the highest, middle and lowest test concentrations, at a minimum. DO and temperature were measured with a YSI dissolved oxygen meter and combination oxygen-temperature probe; pH was measured with an Instrumentation Laboratory portable pH meter and probe.

Results of the definitive tests for fish and water flea were

expressed as the time-depended (24-, 48- and 96-hour and 24-and 48-hour) LC50's, respectively. The LC50 is the nominal concentration of test compound in diluent water which caused 50% mortality in test animal populations at the stated times of exposure. Test concentrations and corresponding observed percentage mortalities were converted to logarithms and probits, respectively, and these values were utilized in a least squares regression analysis. The LC50's and their 95% confidence intervals were calculated from the regression equation.

RESULTS

The pH and DO ranges of selected concentrations during the 96 hours of exposure with fish and 48 hours with water flea are presented in Table 1. The estimated LC50 values, 95% confidence intervals and no discernible effect concentrations for bluegill, channel catfish and fathead minnow are presented in Table 2. Table 3 presents the estimated LC50 values, 95% confidence intervals and no discernible effect concentration for water flea. Table 4 presents a summary of observed percentage mortality for fish in each individual test concentration after 24-, 48- and 96-hour exposures. The observed average percentage mortality for water flea in each individual test concentration after 24- and 48-hour exposure are presented in Table 5. The mortality synárome among fish from those con-

centrations in which mortality was observed was similar. Fish generally became dark and lethargic, lost equilibrium, and expired. All dead water flea observed during this exposure were carrying a partially attached carapace, thus suggesting the organisms died while attempting to molt.

LITERATURE CITED

U.S. EPA. 1975. Methods for Acute Toxicity Tests with

Fish, Macroinvertebrates, and Amphibians by the

Committee on Methods for Toxicity Tests with Aquatic

Organisms. Ecological Research Series (EPA-660/3-75
009, April, 1975), 61 pp.

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Table 1 -- pH and DO ranges of test solutions during exposure

of bluegill (Lepomis macrochirus), channel catfish

(Ictalurus punctatus), fathead minnow (Pimephales

promelas) and the water flea (Daphnia magna) to

hexachlorocyclopentadiene in static, acute toxicity
tests.

Species	рН	DO (mg/l)	
bluegill	7.0-6.5	9.2(104) ^a -0.4(5)	
channel catfish	7.1-6.6	8.5(97)-0.7(8)	
fathead minnow	7.1-6.6	9.2(104)-0.5(6)	
water flea	7.5-6.9	8.0(91)-7.0(80)	

a tof saturation at 22°C.

The state of the s

Table 2 -- Acute toxicity of hexachlorocyclopentadiene to bluegill^a (Lepomis macrochirus) channel catfish^b (Ictalurus punctatus) and fathead minnow^c (Pimephales promelas)

The data are based on results of toxicity tests conducted at the Aquatic Toxicology Laboratory of E G & C, Bionomics, Wareham, Massachusetts.

	LC50	No discernible effect concentration			
Species	24-hour	60 (milligrams/liter) 48-hour 96-hour		through 96 hours (mg/L)	
bluegill	0.17 (0.14-0.21) ^d	0.15 (0.12-0.18)	0.13 (0.11-0.17)	0.065	
channel catfish	0.19 (0.14-0.25)	0.15 (0.13-0.18)	0.097 (0.081-0.12)	0.056	
fathead minnow	0.24 (0.17-0.32)	0.21 (0.18-0.25)	0.18 (0.16-0.22)	0.087	

Toxicity test conducted at $22 \pm 1.0^{\circ}$ C, mean and standard deviation (N=30) wet weight of bluegill was 0.45 \pm 0.17 g.

Toxicity test conducted at 22 \pm 1.0°C, mean and standard deviation (N=30) wet weight of channel catfish was 2.1 \pm 1.0 g.

Toxicity test conducted at 22 \pm 1.0°C, mean and standard deviation (N=30) wet weight of fathcad minnow was 0.72 \pm 0.3 g.

^{95%} confidence interval.

Table 3 -- Acute toxicity of hexachlorocyclopentadiene to the

water flea (Daphnia magna). These data are based

on results of a toxicity test conducted at the Aquatic

Toxicology Laboratory of E G & G, Bionomics, Warcham,

Massachusetts.

LC50 (milligra		No discernible effect concentration through 48 hours
24-hour	48-hour	(mg/l)
0.13 (0.068-0.26) ^b	0.039 (0.030-0.052)	0.018

Toxicity test conducted at 22 \pm 1.0°C, water flea \leq 24 hours old at initiation of the test.

p 95% confidence interval.

Table 4 -- Concentrations tested and corresponding observed percentage mortalities for bluegill (Lepomis macrochirus),
channel catfish (Ictalurus punctatus) and fathead
minnow (Pimephales promelas) exposed to hexachlorocyclopentadiene for 24, 48 and 96 hours.

	Nominal concentration		* mortality observed		
Species	(mg/l)	24-hour		48-hour	96-hour
bluegill	0.32		100	100	100
	0.24		80ª,b	100	100
	0.18		30a,b	30 ^{a,b}	40 ^b
	0.14		10 ^{a,b}	30 ^{a,b}	40 ^b
	0.10		10	10	50 p
	0.087		0	0	10
	0.065		0	0	0
	control	(aceton	e) O	0	0
	control		0	0	0
channel catfish	0.24		100	100	100
	0.18		10 ^b	70 ^C	100
	0.14		o _p	40 ^b	90 ^b
	0.10		0	0	20
	0.075		0	0 _p	30 ^b
	0.056		0	0	0
	control	(aceton	e) û	0	0
	control		0	0	0

Table 4 -- Continued.

	Nominal concentration	<pre>% mortality observed</pre>		
Species	(mg/l)	24-hour	48-hour	96-hour
fathead minnow	0.32	100	100	100
	0.24	50 ^{a,b,c}	80 ^{b,c}	90 ^a ,
	0.18	0	20 ^a	20 ^a
	0.16	0	0	20 ^a
	0.14	0	0	0
	0.12	0	0	10
	0.087	0	0	0
	control (acetone) 0	C	0
	control	0	0	0

a Fish displayed a dark coloration.

b Fish were lethargic.

c Fish displayed a partial loss of equilibrium.

Table 5 -- Concentrations tested and corresponding average observed percentage mortalities for the water flea

(Daphnia magna) exposed to hexachlorocyclopentadiene.

Each mortality value represents the average of 3 replicates.

Nominal concentration	Average percentage mortalit			
(mg/l)	24-hour	48-hour		
0.24	87	100		
0.16	33	100		
0.10	33	100		
0.065	40	93		
0.042	7.	57		
0.028	0	20		
0.018	0	0		
0.012	0	0		
control (acetone)		0		
control	0	0		

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